

# HPLC Application

ID No.: 21877

## Human Plasma TMP & Thiamine by Gemini-NX 3u C18 100x3mm with Impact Plate\_Spiked Plasma

**Column:** Gemini® 3 µm NX-C18 110 Å, LC Column 100 x 3 mm, Ea

**Dimensions:** 100 x 3 mm ID

**Order No:** 00D-4453-Y0

**Elution Type:** Gradient

**Eluent A:** 25nM Na<sub>2</sub>HPO<sub>4</sub>, 10% methanol

**Eluent B:** 25mM Na<sub>2</sub>HPO<sub>4</sub>, 70% methanol

Gradient Profile:	Step No.	Time (min)	Pct A	Pct B
	1	0	3	97
	2	0.25	25	75
	3	0.75	25	75
	4	3	35	65
	5	4	100	0
	6	5	100	0
	7	5.1	3	97
	8	8	3	97

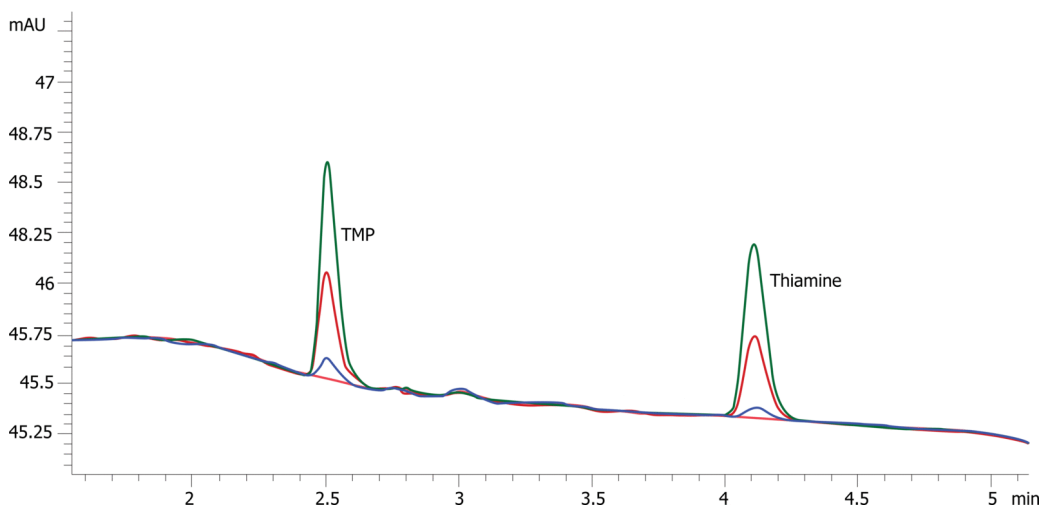
**Flow Rate:** 0.75 mL/min

**Col. Temp.:** ambient

**Detection:** Fluorescence (FLUOR) @ Ex 375 nm (ambient)

**Detector Info:** Shimadzu ELSD

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### ANALYTES:

- 1 TMP  
Retention Time: 2.517 min
- 2 Thiamine  
Retention Time: 4.128 min



Products used in this application:



# Sample Preparation Details

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## Human Plasma TMP & Thiamine by Gemini-NX 3u C18 100x3mm with Impact Plate\_Spiked Plasma

### PRODUCT DESCRIPTION:

Impact<sup>™</sup> Protein Precipitation, 2mL Square Well Filter Plate, 2/Pk

Order No.: CE0-7565

### SOLID PHASE EXTRACTION (SPE) PROCEDURE:

**Note:** The solvent volumes shown below are for a Proprietary bed mass.

The solvent volumes will need to be adjusted for a smaller or larger bed mass.

#### Condition:

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#### Load:

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#### Wash:

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#### Dry:

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#### Elute:

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#### Final Prep and Analysis:

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Inject: 20 µL on HPLC Fluorescence (FLUOR) @ Ex 375 nm (ambient)

<b>ANALYTES:</b>	<b>Spiked Conc. (ng/mL)</b>	<b>Log P</b>	<b>pKa</b>	<b>% Rec</b>	<b>%RSC (n=0)</b>
1 TMP	200				
2 Thiamine	200				

**Note:** This method is designed as a convenient starting point for further investigation and can be tailored to meet your extraction goals. Call your local Phenomenex Representative for assistance in method development and optimization techniques.

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